ELECTROLYTE AND NITROGEN DISTRIBUTION IN WHOLE FAT-FREE SKIN AND HEAT-SEPARATED CORIUM AND EPIDERMIS*†

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After our studies on the distribution of water, nitrogen and electrolytes in the whole skin, it was thought that the different layers of the skin should be investigated separately. The epidermis was therefore separated from the corium and the nitrogen and electrolyte distribution was determined in each of the layers

In view of the fact that adequate literature on the methods for the separation of epidermis from dermis exists in the paper by Baumberger, Suntzeff and Cowdry (1), no formal review of the earlier work will be given.

In choosing a method for the separation of epidermis from corium for chemical analyses, we have to be guided by many considerations such as loss of water; chemical change within the epidermis or dermis during the separation; or shifting of water and electrolytes from one layer to the other, previous to and during the separation.

The method of separation that we used is not entirely satisfactory. In addition to the loss of water during the separation, there is a possibility of chemical change within the epidermis during the separation.

The analytical data as well as the derived data presented here show that the corium layer of the skin has a chemical composition differing so radically from the epidermis that, when analyzed together, as they must be in whole skin analyses, the changes in the corium may mask changes or lack of changes in the epidermis.

This report supplies chemical data on the whole skin, corium and epidermis. These data have been interpreted histochemically and are believed to provide at least a first approximation of (1) the relative mass of the two layers in the whole skin; (2) the amount of fibrillary material in each of the layers; and (3) the distribution of electrolytes in the layers.

PROCEDURE

Normal dogs were kept for 4 weeks in metabolism cages on alternate meat and Globe Dog Chow Blox diet. For the removal of the skin, the dogs were anesthetized with nembutal, given intravenously. The chest and abdomen were clipped with electric clippers and then shaved with a straight edged razor. The shaved skin was washed with distilled water and then dried with gauze. For the whole skin analyses, a strip of skin (10 x 10 cm.) was dissected from the ventral wall on one side of the midline and placed in a glass-stoppered weighing

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bottle. A similar sized strip on the opposite side of the midline was taken and placed in a glass-stoppered weighing bottle to be used for the separation of the epidermis from the corium. Each large piece of skin was placed on a tile and trimmed quickly to remove as much visible free fat as possible. The piece of skin that was to be used for whole skin analysis was cut into small strips approximately 1 x 2 mm. wide and placed in a weighed glass-stoppered weighing bottle, weighed, and placed in a 102° oven and dried to constant weight. The dried skin was then extracted for neutral fat. The strips of fat-free whole skin were transferred to a special apparatus and crushed, using the technic described in previous work (2). The pulverized skin was kept in weighing bottles in a dessicator over activated aluminum oxide. The other large piece of skin was separated into corium and epidermis layers. The dried corium and the dried epidermis layer were each transferred to a special apparatus and crushed, in exactly the same manner as was used for the whole skin sample.

The following determinations were made on the samples of whole dried, crushed skin: total fat, water, chloride, sodium, potassium, calcium, magnesium, total nitrogen and collagen nitrogen (including elastin nitrogen); on the samples of dried crushed corium the same determinations as for whole skin were made, with the exception of water; and on the dried crushed epidermis, fat, chloride, sodium, potassium, total nitrogen and collagen nitrogen (including elastin nitrogen). The number of determinations on the epidermis mass depended upon the amount of tissue available.

SEPARATION OF EPIDERMIS FROM CORIUM

At first it seemed that the ideal method for the separation of epidermis from corium would be by the removal of epidermis on a dermatone directly from the corium from an anesthetized dog, followed by the removal of the corium layer by dissection. Regrettably, this method could not be used by us because the layer of skin that we removed on the dermatone was never thin enough to be only the epidermis. Nevertheless, these two layers of skin so obtained, the one on the dermatone and the dissected layer, were treated in the same way as given previously for whole skin and then analyzed. Along with the separated layers, a sample of whole skin taken from the opposite side of the midline was also analyzed. The data so obtained are presented in table 1.

The points of interest in the data are: the layer of skin removed by dermatone, which should be richer in epidermis than corium, contained less water and more collagen plus elastin nitrogen than the dissected layer of skin (corium), indicating that we removed the upper part of the corium with the epidermis in all of our attempts with the dermatone. It is known (3, 4, 5) that the disposition of the bulk of the elastic fibers are in the upper part of the corium. It is also known that the upper corium is a denser connective tissue containing more fibrillary structures than the lower corium. Therefore, the layer of skin removed on the dermatone not only contained the epidermal system but most of the elastic fibers in the upper part of the corium, as well as some of the collagen of the corium layer.

These results prove that this method, as we had used it, is not satisfactory for separating the epidermal system from the corium.

The method finally adopted for removing epidermis from corium was the heat method of Baumberger, Suntzeff and Cowdry (1). Since water is lost in the dissection of epidermis from the corium by this method, all values for the separated layers had to be expressed in units per 100 gm. of fat-free skin solids. The procedure was as follows: The subcutaneous tissue was trimmed from the large sample of whole skin. The sample was then cut into strips approximately 2 x 12 cm. One strip at a time was then placed with corium layer downward on an asbestos board covered with white oil cloth which was maintained at 50° on the bottom of an electric oven. After a few minutes the epidermis could be pushed away from the dermis layer. The epidermis mass was placed in a weighing bottle for drying to constant weight and fat extraction. The corium layer, after trimming off the outer edges to be sure none of the epidermis layer was left with the corium layer, was further cut into small strips 1 to 2 mm. wide and placed in weighing bottle for drying to constant weight and fat extraction.

TABLE 1

Analyses of whole skin and the layers of skin as removed by dermatone

Units are expressed per 100 gm. of fat-free solids

DOG	TISSUE	H_2O	Cl	Na	K	TOTAL N	COLLAGEN N
		gm.	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*gm.*
SS_1	Whole skin	243	27.1	28.5	6.9	15.4	11.7
	Corium	303	28.4	33.6	11.4	15.0	8.0
	Epidermis	257	32.7			15.3	12.1
SS_3	Whole skin	217	29.3	33.0	5.82	15.5	12.2
	Corium	257	29.3	33.3	5.56	15.6	12.4
	Epidermis	229	28.0			15.3	11.7
SS_4	Whole skin	249	29.1	30.1	6.74	15.9	11.3
	Corium	252	25.9	31.4	8.20	15.4	10.7
	Epidermis	200	23.1	25.4	4.98	16.3	11.6
SS_5	Whole skin	241	27.7	31.4	5.72	16.2	12.9
	Corium	259	27.8	34.8	7.50	15.6	11.8
	Epidermis	206	27.5	26.3	4.15	16.3	12.7
SS_6	Whole skin	222	27.1	30.1	5.56	16.0	12.9
	Corium	217	25.3	28.1	5.49	15.9	12.9
	Epidermis	194	25.5	29.1	5.28	16.5	12.4
SS_7	Whole skin	244	32.6	34.5	7.14	16.1	11.7
	Corium	237	27.7	31.8	8.24	16.1	12.2
	Epidermis	151	26.6			16.3	11.6

RESULTS AND DISCUSSION

The original data for the study consisted of (1) the serum and whole skin values expressed in units per kilo of fat-free whole skin tissue, table 2; (2) the whole skin, corium and epidermis values expressed in units per 100 gm. of fat-free solids, table 3, and (3) the tendon values expressed in units per 100 gm. of fat-free tendon solids, table 4. At the bottom of each of these tables are given the mean values with the standard deviation of the mean.

Since water is lost from the tissue when the layers of skin are separated by heat, all analytical data on corium and epidermis have to be expressed in units per 100 gm. of fat-free solids and thus the derived data have to be expressed in the same manner. From table 3 it will be noted that in the epidermis solids there was found small amounts of what was chemically identified as collagen plus elastin nitrogen (0.16 gm. nitrogen per 100 gm. epidermis solid). Histologically it is known that the collagenous fibers continue upward from the corium into the epidermis in the form of fine fibers. Since the amount of collagen nitrogen determined chemically in epidermis was so small when compared with the collagen values of the corium, we assumed for purpose of calculations that all of the chemically determined collagen nitrogen of whole skin is in the corium layer.

By utilizing the foregoing assumption and applying data obtained from determinations of collagen plus elastin nitrogen and inorganic constituents in whole skin solids and corium solids, it is possible to estimate the proportions of epidermis and corium skin solids in the whole skin solids and also the amounts of the inorganic constituents in these solids. The definitions of the symbols and the equations used in the calculations follow:

(S) = 100 gm. of whole skin solids.

$(S)_c$ = 100 gm. of corium solids.

$(S)_e$ = 100 gm. of epidermis solids.

$(P)_c$ = gm. of corium solids in 100 gm. of whole skin solids (S).

$(P)_e$ = gm. of epidermis solids in 100 gm. of whole skin solids (S).

$(C + E)_s$ = gm. of collagen plus elastin nitrogen in 100 gm. of whole skin solids (S).

$(C + E)_c$ = gm. of collagen plus elastin nitrogen in 100 gm. of corium solids $(S)_c$.

$(C + E)_e$ = gm. of collagen plus elastin nitrogen in 100 gm. of epidermis solids $(S)_e$.

$(Cl)_s$ = m. eq. chloride in 100 gm. of whole skin solids (S).

$(Cl)_{sc}$ = m. eq. chloride in 100 gm. of corium solids $(S)_c$.

$(Cl)_{se}$ = m. eq. chloride in 100 gm. of epidermis solids $(S)_e$.

$(Cl)_{Pc}$ = m. eq. chloride in 89.8 of corium solids.

$(Cl)_{Pe}$ = m. eq. chloride in 10.2 gm. of epidermis solids.

(When constituents other than chloride have been referred to, comparable chemical symbols have been used.)

TABLE 2

Analyses of serum and whole skin of normal dogs

Values are given for fat-free whole skin

DOG NO.	H_2O	Cl	Na	K	Ca	Mg	TOTAL N	COLLAGEN + ELASTIN N
	gm. per Kg.	*m.eq. per Kg.*	*m.eq. per kg.*	*m.eq. per kg.*	*m.eq. per Kg.*	*m.eq. per kg.*	*gm. per Kg.*	*gm. per Kg.*
SS_9								
Serum	917.8	108.0	137.0	4.29			10.07	
Whole skin	705.2	80.40	97.3	29.58			49.2	35.30
SS_{10}								
Serum	927.8	108.5	139.3	4.49			9.16	
Whole skin	745.7	88.7	100.5	23.65			40.75	27.00
SS_{11}								
Serum	914.0	111.6	142.6	4.38	5.80	2.48	10.05	
Whole skin	703.3	86.03	101.2	30.15	6.26	6.70	47.92	32.96
SS_{12}								
Serum	924.1	107.2	139.5	4.18	5.60	2.88	9.33	
Whole skin	740.8	87.4	103.0	24.95	5.60	4.62	42.51	27.96
SS_{13}								
Serum	914.6	107.2	136.9	4.85	5.28	2.08	10.92	
Whole skin	744.5	88.30	102.5	17.92	8.90	4.50	39.90	26.60
SS_{14}								
Serum	917.6	108.0	139.4	4.39	6.14		10.42	
Whole skin	719.1	86.26	90.4	20.81	4.52	4.24	44.97	33.62
SS_{15}								
Serum	922.7	108.2	138.8	4.62	4.60	2.16	9.08	
Whole skin	713.0	80.38	94.6	32.00	7.72	7.26	46.35	30.99
SS_{16}								
Serum	916.2	106.3	137.6	4.46	4.54	2.52	10.22	
Whole skin	684.0	79.90	87.8	23.60	4.86	5.18	51.60	39.90
SS_{17}								
Serum	907.0	118.7	138.0	4.51	5.10	2.04	9.62	
Whole skin	682.5	89.50	90.0	21.52	6.10	5.74	52.0	35.54
Average								
Serum	918.0	109.3	138.8	4.46	5.29	2.36	9.87	
σ*	5.8	3.6	1.6	0.18	0.56	0.09	0.59	
Whole skin	715.3	85.21	96.4	24.91	6.28	5.46	46.13	32.21
σ*	23.0	3.67	5.5	4.48	1.44	1.07	4.21	4.21

* = Standard deviation.

The equations used in the calculation of per cent of corium solids $(P)_c$ and epidermis solids $(P)_e$ in 100 gm. of whole skin solids (S).

TABLE 3

Analyses of whole skin solids and corium and epidermis solids separated by heat (50° C.). Original data

Units are expressed per 100 gm. of fat-free solids

DOG NO.	TISSUE	Cl	Na	K	Ca	Mg	TOTAL N	COLLAGEN + ELASTIN N
		m.eq.	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*gm.*
SS_9	Whole skin	27.27	32.99	7.36			16.7	12.00
	Corium	30.10	31.30	7.72			16.3	13.80
	Epidermis						15.4	0.40
SS_{10}	Whole skin	34.85	39.50	9.29			16.0	10.60
	Corium	33.34	38.92	8.37			16.2	12.00
	Epidermis						15.2	0.20
SS_{11}	Whole skin	29.08	34.10	10.15	2.14	2.04	16.2	11.10
	Corium	31.53	34.01	9.56	2.14	2.50	16.2	12.37
	Epidermis	26.06	31.00				15.7	
SS_{12}	Whole skin	33.70	39.70	9.62	2.16	1.78	16.4	10.78
	Corium	33.20	40.50	9.10	1.86	1.62	16.4	12.50
	Epidermis	28.28	31.50				15.4	0.10
SS_{13}	Whole skin	34.57	40.12	7.02	3.48	1.76	15.6	10.41
	Corium	34.34	40.75	5.42	3.30	1.76	15.5	11.77
	Epidermis	25.54	30.50	12.01				
SS_{14}	Whole skin	30.72	32.30	7.41	1.60	1.50	16.0	11.96
	Corium	27.46	31.45	7.21	1.70	1.52	15.9	13.00
	Epidermis						15.1	0.12
SS_{15}	Whole skin	28.12	33.10	11.11	2.68	2.52	16.1	10.79
	Corium	29.92	34.70	10.66	3.40	2.78	16.4	11.82
	Epidermis	29.28	30.25	12.90				
SS_{16}	Whole skin	25.23	27.75	7.47	1.54	1.64	16.3	12.64
	Corium	25.88	28.40	7.40	2.36	2.16	16.2	13.54
	Epidermis	23.25	31.35	12.00			15.4	0.08
SS_{17}	Whole skin	28.48	28.35	6.78	1.92	1.80	16.3	11.6
	Corium	30.54	31.60	7.75	1.92	1.90	16.4	12.7
	Epidermis	28.67	31.70	11.90			15.3	0.06
Averages	Whole skin	30.22	34.21	8.47	2.22	1.86	16.1	11.32
	σ*	3.23	4.42	1.50	0.66	0.35	0.4	0.72
	Corium	30.70	34.62	8.13	2.38	2.03	16.2	12.61
	σ*	2.62	3.95	1.44	0.64	0.43	0.3	0.71
	Epidermis	26.83	31.02	12.20			15.30	0.16
	σ*	2.21	0.51	0.41			0.17	

* = Standard deviation.

Equations 1

$$(P)_c = \frac{(C + E)_s}{(C + E)_c} \times 100$$

Therefore the grams of epidermis solids $(P)_e$ in (S)

$$(P)_e = 100 - (P)_c$$

To calculate the amounts of inorganic constituents in 100 gm. of epidermis solids $(S)_e$. For example, for chloride

Equations 2

$$(Cl)_{Pc} = \frac{(Cl)_{sc} \times (P)_c}{100}$$

$$(Cl)_{Pe} = (Cl)_S - (Cl)_{Pc}$$

$$(Cl)_{se} = \frac{(Cl)_{Pe} \times 100}{(P)_e}$$

TABLE 4

Average analyses of Achilles, flexor and extensor tendons

Units are expressed per 100 gm. of tendon solids (fat-free)

DOG NO.	WATER	Cl	TOTAL N	COLLAGEN N
	gm.	*m.eq.*	*gm.*	*gm.*
SS_9	162.7	24.04	17.20	15.35
SS_{10}	167.3	24.85	16.75	15.72
SS_{11}	159.2	21.90	17.50	15.85
SS_{12}	151.7	21.06	17.95	16.40
SS_{13}	169.5	20.82	16.42	15.33
SS_{14}	157.6	21.11	17.82	14.54
SS_{15}	150.0	17.52	16.78	15.70
SS_{16}	166.5	19.50	17.26	16.15
Average	160.6	21.35	17.21	15.63
σ*	6.6	2.18	0.68	0.37

* = Standard deviation.

The average findings of this group of dogs (table 3) shows that 100 gm. of whole skin solids (S) contains 11.32 gm. of collagen plus elastin nitrogen, while the 100 gm. of corium solids $(S)_c$ contains 12.61 gm. of collagen plus elastin nitrogen. The 11.32 gm. of collagen nitrogen in the whole skin therefore represents 89.8 gm. of corium solids. Assuming that all of the collagen nitrogen is in the corium layer, the per cent of corium and epidermis in the whole skin solids (S) of the group of 9 dogs was estimated by equations (1) and is given in table 5 and shown graphically in figure 1. Quantitatively, 89.8 per cent of whole skin fat-free solids are corium solids and 10.2 per cent of the whole skin solids

TABLE 5

The percentage of corium and epidermis in 100 gm. of whole fat-free skin solids. (Derived data)

DOG NO.	CORIUM	EPIDERMIS
	per cent	*per cent*
SS_9	87.1	12.9
SS_{10}	88.4	11.6
SS_{11}	89.7	10.3
SS_{12}	86.3	13.7
SS_{13}	88.8	11.2
SS_{14}	92.1	7.9
SS_{15}	91.3	8.7
SS_{16}	93.4	6.6
SS_{17}	91.3	8.7
Average	89.8	10.2
σ*	2.13	2.1

* Standard deviation.

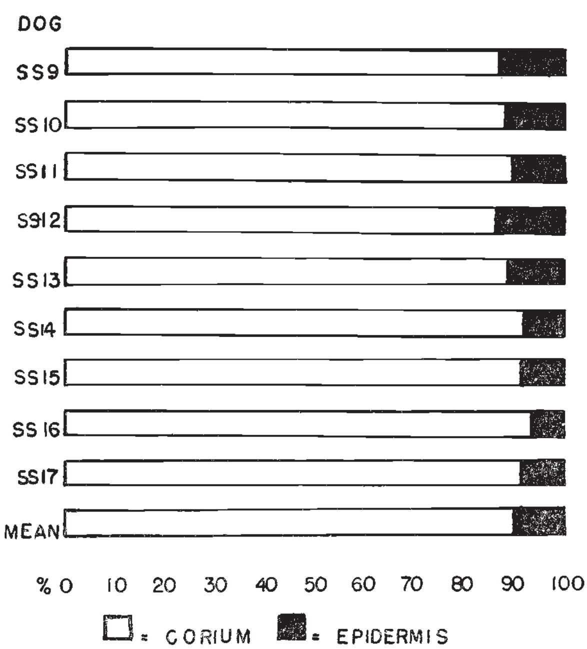

FIG. 1. Graphic representation of the percentage of corium and epidermis in 100 gm. whole fat-free skin solids. The percentage of epidermis is represented by the shaded areas and the corium by the unshaded areas.

are epidermis solids. Using the 10.2 per cent for epidermis, the calculated amounts of electrolytes and total nitrogen in 100 gm. of epidermis solids were calculated (equations 2) from the average whole skin and corium analyses given in table 3. The derived results for 100 gm. of epidermis solids are presented in table 6 along with the original data obtained on the whole skin, corium and epidermis solids. It will be noted that the derived data agrees with the original average analytical data. This method of deriving the electrolyte values in epidermis from simultaneous whole skin and corium analyses should be of value when the amount of epidermis sample is too small for chemical analyses.

The average values obtained on 100 gm. of epidermis solids, original or derived, show that the calcium values (0.88 m. eq.) and magnesium values (0.39) are low when compared with the corium values of 2.38 m. eq. for Ca and 2.03 m. eq. for Mg per 100 gm. of solids. The potassium values in the epidermis of 10.45 m. eq. are higher than the 8.13 m. eq. in the corium solids.

Comparable values are even more clearly shown if the percentage distribution of the electrolytes in the 100 gm. fat-free solids are considered. That is, from the data in table 6 the total base content (Na + K + Ca + Mg) of the 100 gm. of corium solids adds up to a total of 47.16 m. eq. The sodium content of

TABLE 6

Average original data for 100 gm of dry whole skin, corium and epidermis solids and the derived data for 100 gm epidermis solids

(Original data (9 dogs)

	Cl	Na	K	Ca	Mg	TOTAL N	COLLAGEN + ELASTIN N
	m.eq.	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*gm.*
Whole skin	30.22	34.21	8.47	2.22	1.86	16.07	11.32
Corium	30.70	34.62	8.13	2.38	2.03	16.16	12.61
Epidermis	26.83	31.02	12.20			15.30	0.16
Derived data							
Epidermis	26.20	30.50	10.45	0.88	0.39	15.18	0.00

34.62 m. eq. represents 73.6 per cent of the total base amount, K represents 17.2 per cent; calcium, 5.0 per cent and magnesium 4.3 per cent. In 100 gm. of fat-free epidermis solids, the total base content adds up to 42.22 m. eq. of which sodium represents 72.2 per cent; K is 24.7 per cent; calcium is 2.0 per cent and magnesium is 1.0 per cent. Figure 2 shows this percentage distribution graphically.

The following relationships are obvious from the graph: sodium is the predominant cation and chloride is the predominant anion in both layers of skin; the percentage of potassium is higher in epidermis tissue than in corium tissue. In corium the percentage is 17; in epidermis it is 24 per cent of the total base. Minimum amounts of calcium and magnesium are in the corium and only trace amounts are in the epidermis. In corium the percentage of calcium is 5.0; in epidermis it is 2.0. In corium the percentage of magnesium is 4.3; in epidermis it is 1.0 per cent of the total base.

Suntzeff and Carruthers have reported analyses of wet human epidermis

(6) and wet mouse epidermis (7). In both subjects the predominating cation was found to be potassium. In human wet epidermis 52 per cent of the total base content was reported to be potassium and 33.7 per cent was sodium; in wet mouse epidermis 44.5 per cent of the total base was K and 36.5 per cent was Na. These relationships are different from those of ours on dog skin. The reason for this is unknown; however, the techniques of separating the epidermis from the dermis may be one of the reasons; different methods of analyses used may be another; and finally, species difference must be considered.

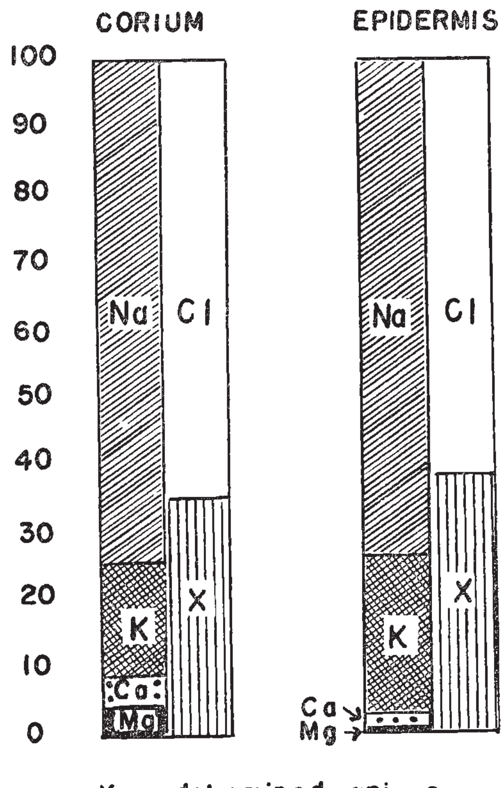

FIG. 2. Graphic representation of the percentage distribution of minerals in 100 gm. fat-free solids of corium and epidermis. The first column in each pair presents the cation percentages and the adjoining column the anion percentages.

Almost all of the collagen + elastin nitrogen was found in the corium solids when the heat method for the separation of the epidermis from the corium layer was used. The large amount of collagen and elastin nitrogen in the corium layer signifies a large amount of fibrillary material. Just how much fiber solids the average 12.61 gm. of collagen N + elastin N (table 3) represents can be calculated from the tendon analyses of this group of dogs (table 4). Tendon is considered the densest connective tissue in the body, meaning that it has the most fibrils, the least ground substance, and all fibroblasts are the same. It will be noted that in 100 gm. of fat-free tendon solids, there were 15.63 gm. of collagen + elastin N (table 4). Therefore the average 12.61 gm. of collagen + elastin nitrogen in 100 gm. of corium solids signifies 80.7 gm. of fibrous material solids. Table 7 gives the calculated fiber solids in 100 gm. of corium solids together with the average value and standard deviation. Knowing the amount of fiber solids in the corium solids and assuming that the fibers of skin are associated with the same amounts of Cl, Na and K as found for tendon, we have calculated the amount of Cl, Na and K identified with that amount of fibrous material using the above tendon data (table 4) along with the sodium and potassium data given by Muntwyler et al. (8). These estimated results are given in table 7.

The first column gives the average fiber solids per 100 gm. of corium solids leaving 19.3 gm. of solids in excess of that of the fibers. This is labeled Δ solids. The Cl, Na, and K in excess of that accounted for by the fibrillary material should represent the amounts associated with the remaining 19.3 gm. of corium solids. The amount of potassium associated with the 80.7 gm. of fiber solids is only 1.25 m. eq. leaving 6.88 m. eq. K to be associated with the remaining 19.3 gm. of corium solids. Therefore the fiber phase of the corium layer of skin is

TABLE 7

Calculated amounts of solids, Cl, Na and K in the estimated fiber solids in 100 gm. of corium solids from the average values of 9 normal dog skins

	FIBER SOLIDS	Δ SOLIDS	FIBER Cl	Δ Cl	FIBER Na	Δ Na	FIBER K	Δ K
	gm.	*gm.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*
Mean	80.7	19.3	17.22	17.40	19.20	14.42	1.25	6.88
Standard deviation	5.7							

Δ = total corium concentration minus fiber concentration.

very poor in potassium. There is another phase, however, that is rich in potassium as well as sodium and chloride.

In 1922, Waterman (9) found by the use of the histochemical methods of McCallum (10) that in the epidermis the hair follicles and hair with the exception of hair root papillae were completely void of calcium; only in extremely good preparations was there seen a small zone in the epidermis containing a little calcium. Much potassium was shown in the hair follicles, glands and hairs in the epidermis. Contrariwise, in the corium layer much calcium was shown and only traces of potassium.

In order to compare the histochemical finding of Waterman with our own analytical results we estimated the partition of electrolytes and nitrogen in the whole skin into amounts for each of the two layers of skin.

From the average analytical data on whole skin solids and corium solids given in table 6 and knowing what per cent of whole skin solids is corium and epidermis, we have estimated the partition of electrolytes in 100 gm. of whole fat-free skin solids. We also estimated the partition in 284.7 gm. of whole skin solids which represent an average kilo of wet fat-free whole skin and have assigned the estimated amounts of electrolytes to the corium and epidermis layers of the skin. These estimations are presented in table 8.

It will be noted that whether analytical or derived results are expressed in units per 100 gm. of fat-free corium solids and epidermis solids (table 6) or in units partitioned into the corium and epidermis of dry or wet fat-free skin (table 8), the corium layer contains, as Waterman found, the majority of the calcium and magnesium of the skin. On the other hand, there is a part of the corium layer which was found to contain more than a trace of potassium. This is contrary to Waterman's findings.

It has been thought for a long time that the K/Ca relationship in skin is of importance. It has been postulated by Luithlen (11, 12) and other authors (13) that the higher the K content in the skin in relation to the calcium, the higher the irritability of the skin against inflammatory stimuli. It is interesting to note that in these experiments the K/Ca relationship in whole skin was found

TABLE 8

Estimated partition of electrolytes in the corium and the epidermis layers of 100 gm. of dry whole fat-free skin

	SOLIDS	Cl	Na	K	Ca	Mg	TOTAL N	COLLAGEN + ELASTIN N
	gm.	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*m.eq.*	*gm.*	*gm.*
Whole skin	100.0	30.22	34.21	8.47	2.22	7.86	16.07	11.32
Corium	89.8	27.60	31.10	7.30	2.14	1.67	14.52	11.32
Epidermis	10.2	2.62	3.11	1.17	0.08	0.19	1.55	0.00
Estimated partition of electrolytes in the corium and the epidermis layers of 284.7 gm. of whole fat-free skin solid which represents 1 kilo of wet fat-free skin								
Whole skin	284.7	86.20	97.4	24.10	6.32	5.30	45.75	32.25
Corium	255.7	78.60	88.6	20.78	6.09	4.76	41.15	32.25
Epidermis	29.0	7.60	8.8	3.32	0.23	0.54	4.60	0.00

to be 8.47 m. eq./2.22 m. eq. equals 3.8; for corium 8.13/2.38 equals 3.4; and for epidermis 10.45/0.88 equals 12.0. The whole skin ratio of 3.8 completely masked the ratio in the epidermis which is approximately four times that of the corium. This is proof that for further study on the tissue skin, the layers must be separated and the corium and epidermis changes determined on each layer.

Since the method used here for the separation of epidermis from the dermis prevented the exact determination of water in the separated layers it is imperative that further investigation be done in order to give us a method of separation that will allow the exact distribution of water and minerals in the different layers of skin.

SUMMARY

Whole skin of normal dogs was analyzed for total fat, water, chloride, sodium, potassium, calcium, magnesium, total nitrogen and collagen plus elastin nitrogen.

By use of the heat method of Baumberger, Suntzeff and Cowdry, whole skin was separated into the corium and epidermis layers. The same chemical determinations as for whole skin were made on the corium layer with the exception of water while the number of determinations on the epidermis layer depended upon the amount of tissue available. The data have been used to furnish a histochemical description of the two layers of skin:

1. For 100 gm. of fat-free corium solids from normal dogs the average means were as follows: chloride, 30.70 m. eq.; sodium, 34.62 m. eq.; potassium, 8.13 m. eq.; calcium, 2.38 m. eq.; magnesium, 2.03 m. eq.; total nitrogen, 16.16 gm.; and collagen plus elastin nitrogen, 12.61 gm.
2. For 100 gm. of fat-free epidermis solids, the average means were: chloride, 26.20 m. eq.; sodium, 30.50 m. eq.; potassium, 10.45 m. eq.; calcium, 0.88 m. eq.; magnesium, 0.39 m. eq.; total nitrogen, 15.18 gm.
3. 100 gm. of normal fat-free whole skin solids consists of corium solids amounting to an average of 89.8 per cent and epidermis solids amounting to 10.2 per cent. Such a conclusion involved the assumption that all of the collagen plus elastin nitrogen was in the corium layer. The method used here for the derivation of the electrolyte values in epidermis, from simultaneous whole skin and corium analyses, should be of value when the amount of epidermis sample is too small for chemical analyses.
4. An average value of 80.7 per cent of corium solids are fiber solids. This estimation involved the assumption that the fibrillary material of skin is associated with the same amounts of collagen, water, chloride, etc. as found for the connective tissue, tendon.

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